

INFLUENCE OF TURCICUM LEAF BLIGHT DISEASE ON BIOCHEMICAL CHANGES IN MAIZE IN RELATION TO DISEASE RESISTANCE

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ABSTRACT

*The biochemical changes in resistant and susceptible varieties of maize caused by *Exserohilum turcicum* showed that maximum phenols, sugars, chlorophyll and protein content was observed in resistant varieties MI-12 and MI-14. Sugars, chlorophyll and protein contents were higher in healthy leaves of resistant variety compared to infected leaves but the phenol content was high in infected leaves of resistant varieties due to infection it increased drastically. Phenol content was maximum in MI-14 in infected leaves whereas Sugars, chlorophyll and protein contents were maximum in MI-12 of healthy leaves. The contents of sugars, phenols, chlorophyll and protein were higher at 30 DAS and it was decreased gradually from 60 to 90DAS in all samples irrespective of varieties.*

KEYWORDS: *Exserohilum Turcicum, Phenols, Protein, Sugars and Chlorophyll*

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INTRODUCTION

Maize (*Zea mays* L.) is an important coarse cereal crop of the world. It is cultivated in tropics, sub-tropics and temperate regions under irrigated to rainfed conditions it is widely cultivated throughout the world and a higher quantity of maize is produced each year than any other grain. While the United States produces almost half of the world's harvest (~42.5%). Karnataka state accounts for 1.38 million ha area with a production of 3.98 mt and 2883 kg/ha productivity (Anon, 2014). Among the various constraints, the crop suffering from various diseases is a major setback to the production. Among the diseases turcicum leaf blight (TLB) of maize caused by *Exserohilum turcicum* (Pass.) Leonard and Suggs is an important foliar disease of worldwide importance (Carlos, 1997). More than 50 per cent loss in grain yield was reported in USA (Robert, 1953; Raymundo and Hooker, 1981).

Though, there are fungicides that can reduce disease development, but they are not made economical and causes environmental pollution. Development of resistant varieties is the most appropriate approach to control the diseases. Generally, resistance in plants against various diseases has been correlated with various biochemical substances sugars, proteins, chlorophyll and phenolic compounds. However, information on relationship between resistance to TLB and biochemical parameters in maize is very scanty. Therefore the present investigations were focused to identify biochemical parameters associated with different gradients of maize resistance to TLB.

MATERIALS AND METHODS

The seeds sown in pots with different resistance level in glass house conditions. Seedlings were raised in controlled conditions with recommended package of practices. One set maize inbred lines with different resistance levels were maintained for healthy for entire cropping period. Another set was maintained with artificial inoculation of *E. turcicum* through foliar spray for diseased condition. In each set 10 seedlings were maintained

for each variety. The leaf samples were drawn for biochemical analysis at 30 DAS (days after sowing), 60 DAS and 90DAS. Healthy and diseased leaves from genotypes MI-12, MI-14 resistant, NEI-9202B, NEI-9208B Susceptible and CM-202, highly susceptible to TLB were collected 30 days after inoculation and w subjected for extraction in 80 per cent ethanol separately. The extracts were analysed for total chlorophyll, chlorophyll 'a' and chlorophyll 'b' as per the standard protocols (Arnon, 1949), total phenols (Bray and Thorpe, 1954), Total protein (Lowry *et al.*, 1951) total sugars and reducing sugars (Nelson, 1944). Nonreducing sugars were calculated by subtracting the amount of reducing sugars from total sugars.

RESULTS AND DISCUSSIONS

Analysis of biochemical constituents revealed that, at 30DAS there was variation in phenol content across both the resistant and susceptible maize inbred lines. However, maximum phenols was observed in infected leaves of resistant inbred line MI-14 (3.87 mg/g) followed by MI-12 (3.37 mg/g) when compared to healthy leaves (2.75 and 2.62 mg/g) respectively. In susceptible inbred lines also maximum phenol was observed in infected leaves of NEI-9208B (2.60 mg/g) followed by NEI9202B (2.50 mg/g) compare to healthy leaves (2.12 and 2.05 mg/g) of respective inbred lines. Similarly at 60 DAS, maximum phenols were observed in infected leaves of resistant inbred lines compared to susceptible inbred lines (Table 1). But the maximum increase in phenol content from healthy to infected was found in CM202 with 0.28 to 0.62 mg/g. At 90 DAS, maximum increase in phenol content was observed in MI-14 from healthy to infected (0.71 to 0.98 mg/g) followed by NEI-9202B (0.53 to 0.75 mg/g). The high phenol content in resistant inbred lines may be due to more sugar as it acts as precursor for synthesis of phenolics. Phenol content and its enhancement during disease progress were least in susceptible varieties. The results are in confirmation with the findings of Nagaveni (2005) and Pradeepkumar (2005).

Similarly the variation in protein content was also observed in maize inbred lines with different resistance levels. The results showed that reduction of protein content was observed after pathogen infection but more reduction was recorded in the susceptible inbred lines than that of resistant inbred lines. At 30 DAS protein content was more in healthy samples of both resistant and susceptible inbred lines (7.12 to 8.91 mg/g) than in infected samples (5.56 to 8.08 mg/g).

Table 1: Influence of Turcicum Leaf Blight on Phenol Content (Mg/G) in Different Inbred Lines of Maize

Inbred Lines	Phenols (mg/g)					
	30DAS		60DAS		90DAS	
	Healthy	Infected	Healthy	Infected	Healthy	Infected
NEI-9202B (S)	2.05	2.50	1.06	1.05	0.53	0.75
NEI-9208B (S)	2.12	2.60	0.90	1.15	0.28	0.46
CM-202 (HS)	2.05	2.17	0.28	0.62	0.25	0.35
MI-12 (R)	2.62	3.37	1.71	1.75	0.91	1.01
MI-14 (R)	2.75	3.87	1.28	1.31	0.71	0.98
S.Em±	0.10	0.16	0.07	0.07	0.03	0.02
C.D. at 1%	0.45	0.74	0.31	0.33	0.14	0.10

(R) – Resistant, (S) – Susceptible, (HS) – Highly susceptible

Table 2: Influence of Turcicum Leaf Blight on Soluble Protein Content (Mg/G) in Different Inbred Lines of Maize

Inbred Lines	Soluble Protein (mg/g)					
	30DAS		60DAS		90DAS	
	Healthy	Infected	Healthy	Infected	Healthy	Infected
NEI-9202B (S)	7.90	5.56	7.05	5.08	5.82	4.39

Table 2: Contd.,						
NEI-9208B (S)	7.12	6.32	6.20	5.48	5.41	5.01
CM-202 (HS)	7.95	5.96	6.98	5.12	5.21	4.19
MI-12 (R)	8.91	8.08	8.31	7.85	7.94	7.34
MI-14 (R)	8.58	7.98	7.94	7.54	7.52	6.94
S.Em±	0.10	0.007	0.05	0.01	0.00	0.03
C.D. at 1%	0.47	0.03	0.26	0.05	0.03	0.16

(R) – Resistant, (S) – Susceptible, (HS) – Highly susceptible

However, the maximum protein content was observed in healthy leaves of resistant inbred lines MI-12 (8.91 mg/g) followed by MI-14 (8.58 mg/g). Similarly at 60 and 90 DAS, it was gradually decreased in both diseased and healthy samples. But maximum protein content was observed in healthy leaves of resistant inbred line MI-12 (8.31 mg/g) at 60 and (7.94 mg/g) at 90DAS compared to infected leaves 7.85 mg/g and 7.34 mg/g respectively. In case of five inbred lines with differed in their resistance levels, Protein content was more in resistant inbred lines in both healthy and diseased ones compared to susceptible and highly susceptible inbred lines in 30, 60 and 90DAS (Table 2). The protein content decreased slightly from 30 to 90 DAS. Pradeepkumar (2005) and Nagaveni (2005) also obtained the similar results. In contrary to the present findings Arjunan *et al.* (1976) found more protein nitrogen in infected leaves of sorghum infected by *H. turcicum* compare to healthy leaves. The decrease may be due to degradative activity. The protein biosynthesis of the host is widely assumed to be significant feature of pathogenesis, particularly during incompatible reaction.

At 30DAS, the total chlorophyll was ranged as 2.27 and 1.86 mg/g in MI-12 and 2.66 and 2.06 mg/g in MI-14 in healthy and infected leaves, respectively. In case of susceptible inbred lines, it was 1.60 and 1.30 mg/g in NEI-9202B and 1.88 and 1.14 mg/g in NEI-9209B in healthy and infected leaves, respectively. In highly susceptible inbred line CM-202, it was 1.03 mg/g in healthy and 0.99 mg/g in infected leaf, respectively (Table 3). Later, 60DAS and 90DAS it was gradually decreases in all the inbred lines but maximum decrease were found in susceptible and highly susceptible inbred lines compared to resistant inbred lines. All types of chlorophyll was maximum in resistant inbred lines compared to susceptible inbred lines. It has been observed that there was two to three fold reduction of all the types of chlorophyll from 60DAS to 90DAS. This stage plays a crucial role for yield reduction. Similar results on reduction of chlorophyll content after pathogen infection was obtained by earlier workers (Amaresh, 2000 and Mesta, 2006).

Across the different stages, in resistant inbred lines, total sugar content was ranged from 11.5 to 25.5 mg/g and 9.5 to 20.5 mg/g in healthy and diseased samples, respectively. Whereas in susceptible and highly susceptible inbred lines, it was ranged from 5.00 to 17.50 mg/g and 3.5 to 15 mg/g in healthy and diseased samples respectively. Similarly reducing sugar in resistant inbred lines ranged from 8.22 to 18 mg/g and 2.20 to 22.5 mg/g and in susceptible inbred lines, it was 2.8 to 22.5 mg/g and 2.2 to 9.75 mg/g in healthy and diseased samples, respectively. Non- reducing sugar was ranged from 3.28 to 7.5 mg/g and 2.80 to 5.10 mg/g in resistant whereas in susceptible inbred lines it was 2.2 to 7.2 mg/g and 1.3 to 5.25 mg/g in healthy and diseased samples, respectively.

Maximum total sugar content was observed in resistant inbred line of healthy leaves of MI-12 (25.5 mg/g) and MI-14 (22.5 mg/g) compared to infected leaves 20.5 and 12.5 mg/g respectively. The maximum reducing sugar was observed in MI-12 (18.0 mg/g) followed by MI-14 (17.0 mg/g) in healthy leaves compared to infected leaves (15.4 mg/g) and (8.70 mg/g) respectively (Table 4). The maximum non-reducing sugar was observed in MI-12 (7.50mg/g) followed by MI-14 (5.50 mg/g) in healthy leaves compared to infected leaves (5.10 mg/g) and (3.80 mg/g) respectively. Similar observations were obtained by Nagaveni (2005) and Pradeepkumar (2005) that the high level of sugars in the host plant are

stated to be responsible for disease resistance. It was less in infected leaves as compared to healthy leaves (Sandeep *et al.*, 2009). Since *E. turcicum* is high sugar pathogen, during the infection utilize the sugar for growth and development. Therefore, the sugar content was less in susceptible lines as well as in infected leaves as compare to healthy leaves.

CONCLUSIONS

Phenol content increased after pathogen infection but more phenol accumulation was found in diseased samples than that of healthy leaves. At 30 days after sowing phenol content in diseased leaf sample was more and it was decreased in case of healthy sample,

Table 3: Effect of Turcicum Leaf Blight on Chlorophyll Content (Mg/G) in Inbred Lines of Maize with Different Levels of Resistance

Inbred lines	Chlorophyll Content (mg/g)																	
	30 DAS						60 DAS						90 DAS					
	Healthy			Infected			Healthy			Infected			Healthy			Infected		
	Chl. 'a'	Chl. 'b'	Total chloro-phyll	Chl. 'a'	Chl. 'b'	Total chloro-phyll	Chl. 'a'	Chl. 'b'	Total chloro-phyll	Chl. 'a'	Chl. 'b'	Total chloro-phyll	Chl. 'a'	Chl. 'b'	Total chloro-phyll	Chl. 'a'	Chl. 'b'	Total chloro-phyll
NEI-9202B (S)	1.18	0.42	1.60	0.98	0.32	1.30	1.04	0.32	1.36	0.91	0.27	1.18	0.57	0.58	1.15	0.29	0.19	0.48
NEI-9208B (S)	1.40	0.48	1.88	0.82	0.32	1.14	1.34	0.40	1.74	0.75	0.22	0.97	0.92	0.27	1.19	0.39	0.05	0.44
CM-202 (HS)	0.81	0.22	1.03	0.78	0.21	0.99	0.79	0.18	0.97	0.74	0.17	0.91	0.73	0.08	0.81	0.21	0.07	0.28
MI-12 (R)	1.75	0.52	2.27	1.38	0.48	1.86	1.50	0.48	1.98	1.20	0.35	1.55	0.98	0.34	1.34	0.47	0.16	0.63
MI-14 (R)	1.88	0.78	2.66	1.08	0.98	2.06	1.63	0.69	2.32	0.93	0.85	1.78	1.26	0.40	1.64	0.56	0.19	0.75
S.Em±	0.026	0.006	0.026	0.006	0.005	0.026	0.052	0.058	0.045	0.051	0.017	0.015	0.013	0.052	0.047	0.007	0.017	0.065
C.D. at 1%	0.118	0.026	0.118	0.026	0.023	0.118	0.232	0.259	0.200	0.227	0.078	0.068	0.059	0.234	0.209	0.033	0.078	0.293

(R) – Resistant, (S) – Susceptible, (HS) – Highly susceptible

Table 4: Influence of Turcicum Leaf Blight on Sugar Content (Mg/G of Fresh Weight) in Different Inbred Lines of Maize

Data-Tion	Inbred Lines	Sugars (mg/g)										S.Em±		C.D. at 1%	
		NEI-9202B (S)		NEI-9208B (S)		CM-202 (HS)		MI-12 (R)		MI-14 (R)		Healthy	Infected	Healthy	Infected
		Healthy	Infected	Healthy	Infected	Healthy	Infected	Healthy	Infected	Healthy	Infected				
30 DAS	Total sugars	17.0	15.0	13.0	12.2	17.5	13.7	25.5	20.5	22.5	12.5	0.42	0.95	1.91	2.25
	Reducing sugars	9.80	9.75	7.30	9.40	22.5	8.00	18.0	15.4	17.0	8.70	0.44	0.398	1.99	1.78
	Non-reducing sugars	7.20	5.25	5.70	2.80	5.00	5.70	7.50	5.10	5.50	3.80	0.33	0.29	1.47	1.33
60 DAS	Total sugars	13.5	11.0	12.3	7.50	10.5	5.70	18.5	15.0	12.0	11.5	0.56	0.65	2.54	2.91
	Reducing sugars	7.50	7.20	5.80	5.50	3.50	3.00	12.0	10.3	8.25	7.80	0.40	0.36	1.79	1.63
	Non-reducing sugars	6.00	3.80	5.50	2.00	4.00	2.70	6.50	4.70	3.75	3.20	0.78	0.42	3.53	1.90
90 DAS	Total sugars	7.50	7.49	8.50	6.56	5.00	3.50	15.0	13.0	11.5	9.50	0.84	0.70	3.79	3.17
	Reducing sugars	5.00	5.50	6.20	4.40	2.80	2.20	11.5	9.20	8.22	6.64	0.47	0.18	2.11	0.81
	Non-reducing sugars	2.50	1.99	2.30	2.16	2.20	1.30	3.50	2.80	3.28	2.86	0.06	0.06	0.30	0.29

(R) – Resistant, (S) – Susceptible, (HS) – Highly susceptible

Whereas at 60 and 90 DAS it was gradually decreasing in both diseased as well as healthy samples. Maximum protein content was observed in healthy leaves of resistant variety. Protein content was more in resistant varieties in both

healthy and diseased ones compared to susceptible and highly susceptible varieties in 30, 60 and 90DAS. Total sugar, reducing sugar and non-reducing sugar content was less in diseased samples when compared to healthy samples in both resistant and susceptible varieties. At 30 DAS, sugar content was more in healthy leaf samples whereas diseased leaf samples recorded less sugar content. At 60 and 90 DAS, sugar content was gradually decreases in both healthy and diseased samples whereas chlorophyll contents were less in diseased samples compared to healthy samples irrespective of varieties. All the biochemical constituents estimated except phenols were less in infected leaves compare to healthy leaves later on gradually reduced. These constituents plays significant role in imparting resistance in maize against turcicum leaf blight.

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